

Supplementary Material

Tab. S1 - The methods used for determining soil parameters.

Parameter	Method
Soil bulk density (BD)	Obtained from each soil layer using the cutting ring method (50 mm diameter, 50 mm height)
Soil organic carbon (SOC)	Determined using an elemental analyzer (Elementar total organic carbon/total nitrogen analyzer; Germany) according to Bao (2000)
Soil inorganic carbon (SIC)	SIC content was analyzed using the HCl method, through manometric collection of CO ₂ evolved after adding excessive volumes of 4 M HCl into the soil samples (Calcimeter, Eijkelkamp, Netherlands) .
Total nitrogen content (TN)	Determined through sulfuric acid digestion and using FOSS automatic nitrogen determination apparatus according to Bao (2000)
Available nitrogen content (AN)	Analyzed by the Alkali diffusion method according to Bao (2000)
Total phosphorus content (TP)	Determined by NaOH digestion followed by ammonium-molybdate colorimetry (T181 XinShiJi ultraviolet spectrophotometer) according to Bao (2000)
Available phosphorus content (AP)	Extracted using the NaHCO ₃ -ultraviolet spectrometer subsystem (T181 XinShiJi ultraviolet spectrophotometer) according to Bao (2000)
Available potassium (AK)	Extracted with ammonium acetate according to Bao (2000)
Available calcium (ACa) and available magnesium (AMg)	Determined using a soil: water (1: 5) mixture by an Atomic Absorption Spectrophotometer according to Bao (2000)
β-glucosidase	According to “Instructions for the use of the kit” from Shanghai Enzyme-linked Biotechnology Co., Ltd., China, the assay procedure are as follows: <ol style="list-style-type: none"> 1. Add standard: Set Standard wells, testing sample wells. Add standard 50 μl to standard well. 2. Add sample: Set blank wells separately (blank comparison wells don't add sample and HRP-Conjugate reagent, other each step operation is same). Testing sample well. Add Sample dilution 40 μl to testing sample well, then add testing sample 10 μl (sample final dilution is 5-fold), add sample to wells, don't touch the well wall as far as possible, and Gently mix. 3. Add enzyme: Add HRP-Conjugate reagent 100 μl to each well, except blank well. 4. Incubate: After closing plate with Closure plate membrane, incubate for 60 min at 37 °C. 5. Configurate liquid: 20-fold wash solution diluted 20-fold with distilled water and reserve. 6. Washing: Uncover Closure plate membrane, discard Liquid, dry by swing, add washing buffer to every well, still for 30 s then drain, repeat 5 times, dry by pat. 7. Color: Add Chromogen Solution A 50 ul and Chromogen Solution B to each well, evade the light preservation for 15 min at 37 °C 8. Stop the reaction: Add Stop Solution 50 μl to each well, Stop the reaction (the blue color change to yellow color). 9. Assay: take blank well as zero, Read absorbance at 450 nm after Adding Stop Solution and within 15min.
Leucine aminopeptidase	According to “Instructions for the use of the kit” from Shanghai Enzyme-linked Biotechnology Co., Ltd., China, the assay procedure are as follows: <ol style="list-style-type: none"> 1. Add standard: Set Standard wells, testing sample wells. Add

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N-acetyl glucose aminidase	<p>According to "Instructions for the use of the kit" from Shanghai Enzyme-linked Biotechnology Co., Ltd., China, the assay procedure are as follows:</p> <ol style="list-style-type: none"> Add standard: Set Standard wells, testing sample wells. Add standard 50 μl to standard well. Add sample: Set blank wells separately (blank comparison wells don't add sample and HRP-Conjugate reagent, other each step operation is same). Testing sample well. add Sample dilution 40 μl to testing sample well, then add testing sample 10 μl (sample final dilution is 5-fold), add sample to wells, don't touch the well wall as far as possible, and Gently mix. Add enzyme: Add HRP-Conjugate reagent 100 μl to each well, except blank well. Incubate: After closing plate with Closure plate membrane, incubate for 60 min at 37 °C. Configurate liquid: 20-fold wash solution diluted 20-fold with distilled water and reserve. Washing: Uncover Closure plate membrane, discard Liquid, dry by swing, add washing buffer to every well, still for 30 s then drain, repeat 5 times, dry by pat. Color: Add Chromogen Solution A 50 ul and Chromogen Solution B to each well, evade the light preservation for 15 min at 37 °C Stop the reaction: Add Stop Solution 50 μl to each well, Stop the reaction (the blue color change to yellow color). Assay: Take blank well as zero, read absorbance at 450 nm after Adding Stop Solution and within 15 min.
Acid phosphatase	<p>According to "Instructions for the use of the kit" from Shanghai Enzyme-linked Biotechnology Co., Ltd., China, the assay procedure are as follows:</p> <ol style="list-style-type: none"> Add standard: Set Standard wells, testing sample wells. Add standard 50 μl to standard well. Add sample: Set blank wells separately (blank comparison wells don't add sample and HRP-Conjugate reagent, other each step operation is same). Test sample well. Add Sample dilution 40 μl to

Ding L, Wang P, Zhang W, Zhang Y, Li S, Wei X, Chen X, Zhang Y, Yang F (2020).

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Parameter	Method
	testing sample well, then add testing sample 10 μ l (sample final dilution is 5-fold), add sample to wells, don't touch the well wall as far as possible, and Gently mix.
	3. Add enzyme: Add HRP-Conjugate reagent 100 μ l to each well, except blank well.
	4. Incubate: After closing plate with Closure plate membrane, incubate for 60 min at 37 °C.
	5. Configurating liquid: 20-fold wash solution diluted 20-fold with distilled water and reserve.
	6. Washing: Uncover Closure plate membrane, discard Liquid, dry by swing, add washing buffer to every well, still for 30 s then drain, repeat 5 times, dry by pat.
	7. Color: Add Chromogen Solution A 50 μ l and Chromogen Solution B to each well, evade the light preservation for 15 min at 37 °C
	8. Stop the reaction: Add Stop Solution 50 μ l to each well, Stop the reaction (the blue color change to yellow color).
	9. Assay: Take blank well as zero, Read absorbance at 450 nm after Adding Stop Solution and within 15 min.

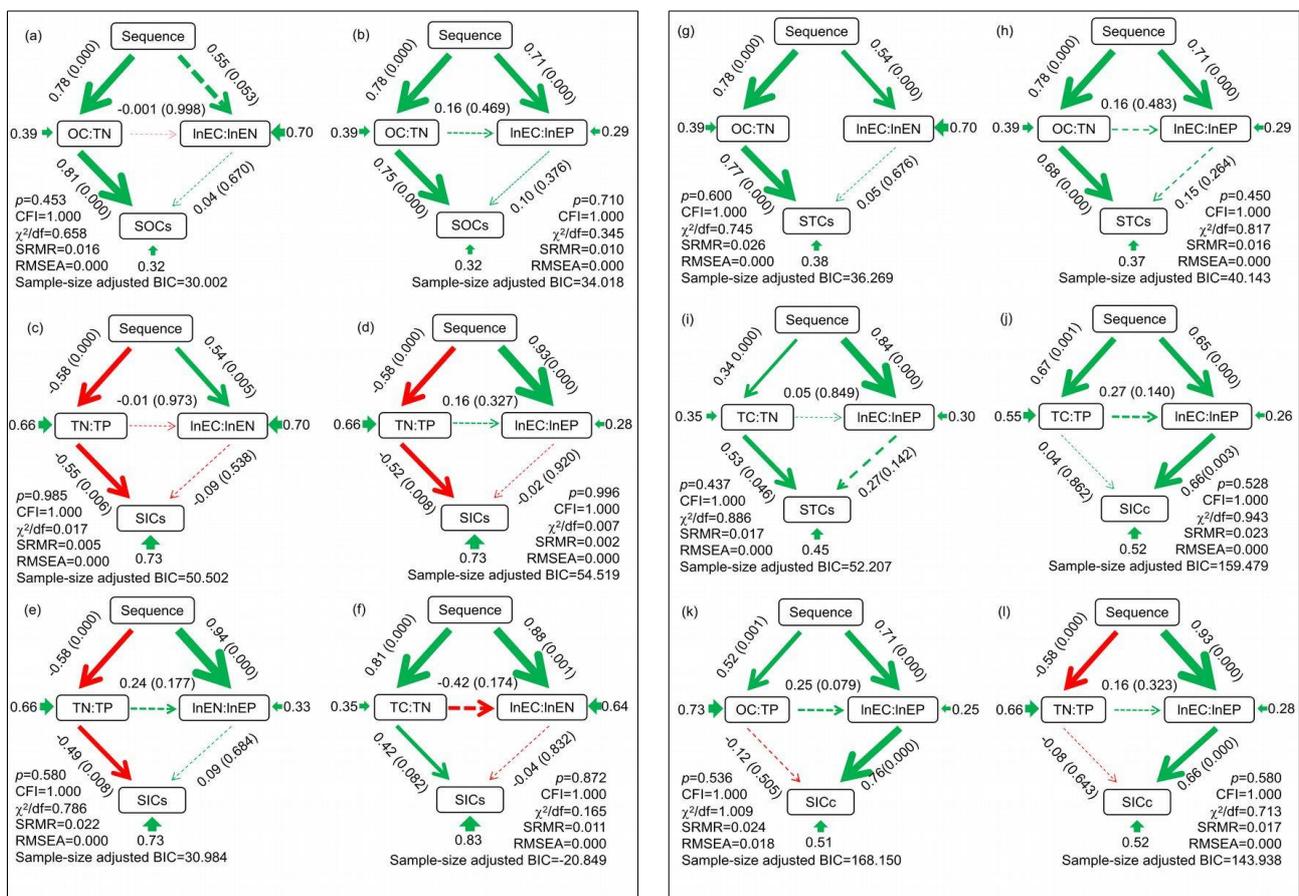
References

- Bao DS (2000). Soil and Agricultural Chemistry Analysis. 3rd ed. China Agriculture Press, Beijing, pp,14-210
- Li C, Li Q, Zhao L, Ge S, Chen D, Dong Q, Zhao X (2016). Land-use effects on organic and inorganic carbon patterns in the topsoil around Qinghai Lake basin, Qinghai-Tibetan Plateau. *Catena* 147: 345-55. -doi: [10.1016/j.catena.2016.07.040](https://doi.org/10.1016/j.catena.2016.07.040)

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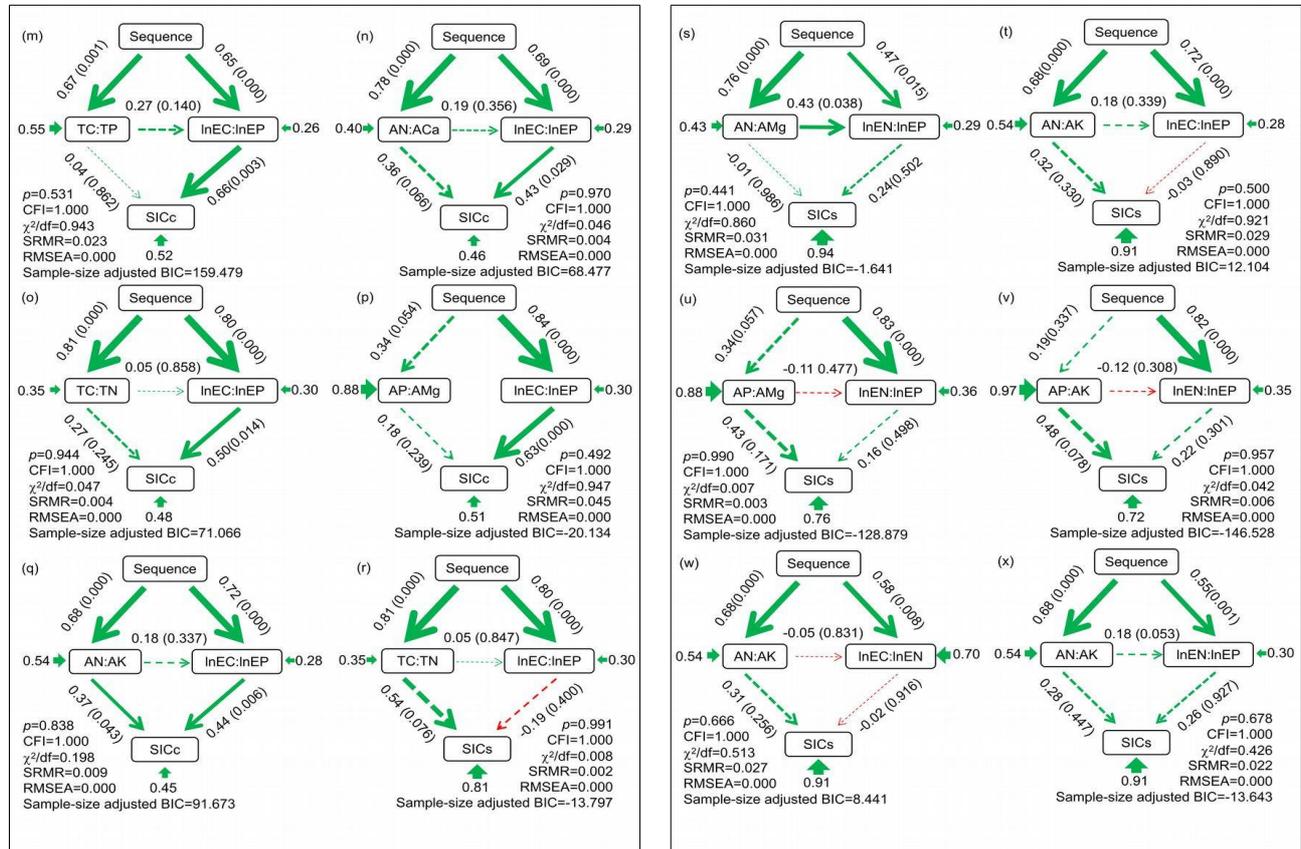
Fig.S1 - Structural equation models reveal the mediating effect of soil stoichiometry. Structural equation models showing SES effects on SOC, SIC and STC concentrations and stock changes through hypothetical pathways of stoichiometry factors. $0 \leq \chi^2/df \leq 2$, $0.05 < p \leq 1$, $0.97 \leq CFI \leq 1$, $0 \leq RMSEA \leq 0.05$ and $0 \leq SRMR \leq 0.05$ indicate good models. CFI = Comparative Fit Index, SRMR = Standardized Root Mean Square Residual, RMSEA = Root Mean Square Error of Approximation. Solid lines show significant pathways ($p < 0.05$), dotted lines indicate non-significant pathways ($p > 0.05$), with line thickness representing the strength (coefficient) of the association between variables; green indicates positive coefficient (r); red indicates negative coefficient (r); and arrows indicate directionality. Alongside the arrow, the numbers outside the brackets are path coefficients; numbers inside the brackets are p values. The numbers at the originating end of the arrow are R^2 for error variables. Sequences indicate the shrub encroachment stages. The abbreviations are listed in the method section of the parent paper.



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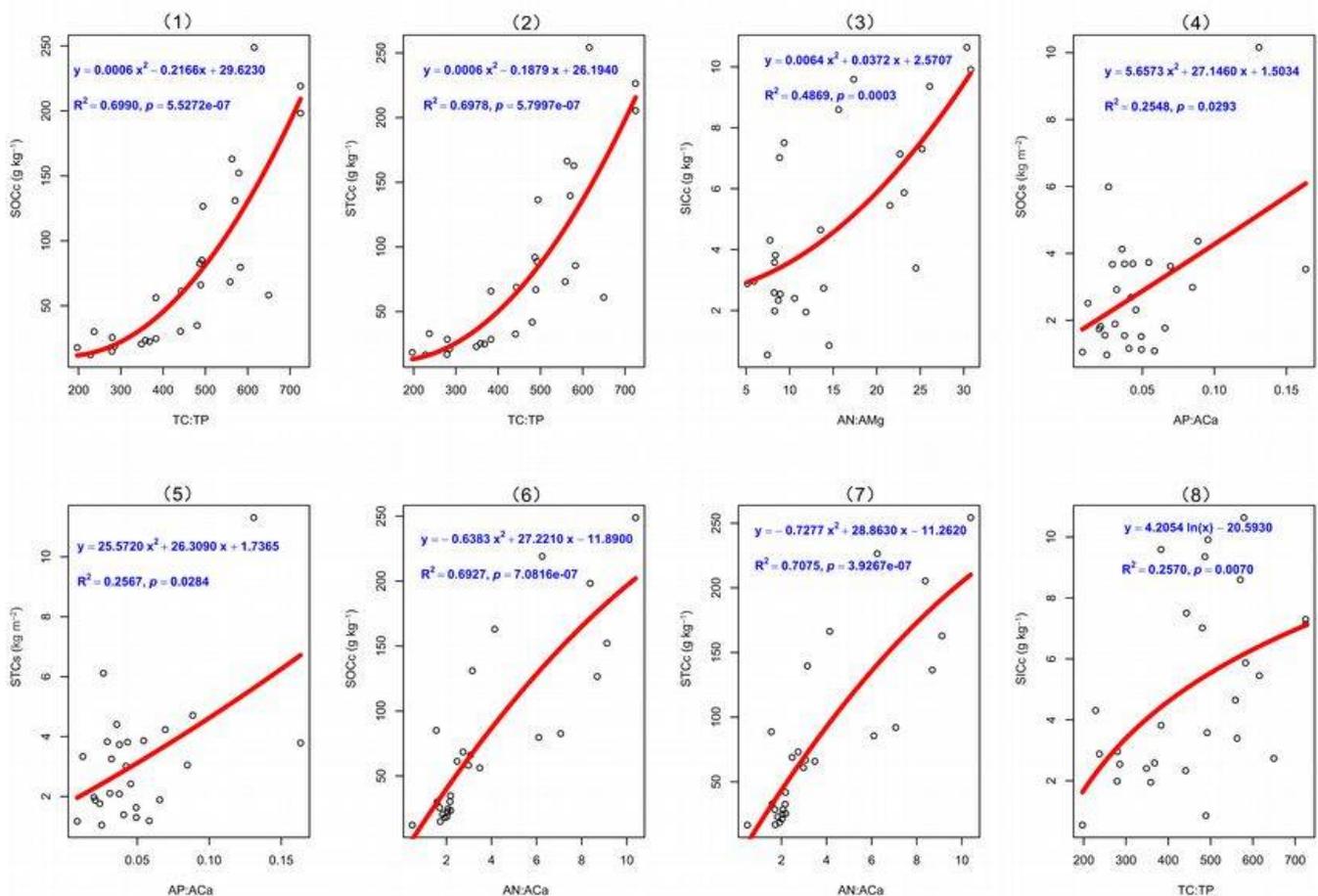
Fig. S1 – (continued).



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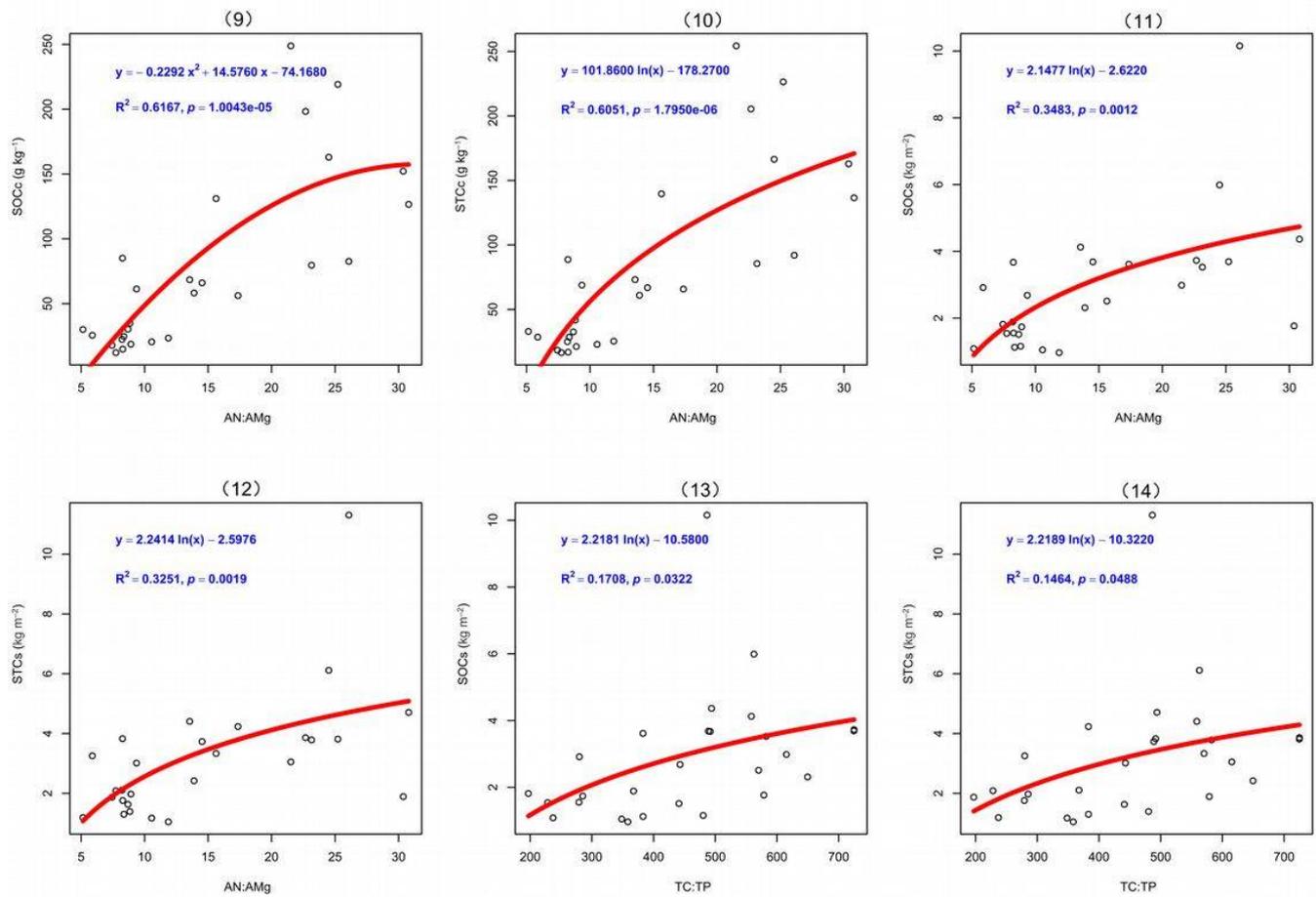
Fig. S2 - Regression analysis reveals soil stoichiometry changes soil carbon (C). E.g., Fig.S2 (1-21) showed that the monotonously increasing or decreasing responses of soil C to soil nutrient stoichiometry ratios, but Fig.S2 (22-36) showed that the threshold responses of soil C to soil nutrient stoichiometry ratios. Fig.S2 (41-49) showed that the monotonously increasing of soil C to soil C-, nitrogen (N)- and phosphorus (P)-acquiring enzymes stoichiometry ratios. The response patterns of soil carbon to soil nutrient stoichiometry ratio showed diversity, while the response to enzyme ratio showed singularity. The abbreviations are listed in the method section of the parent paper.



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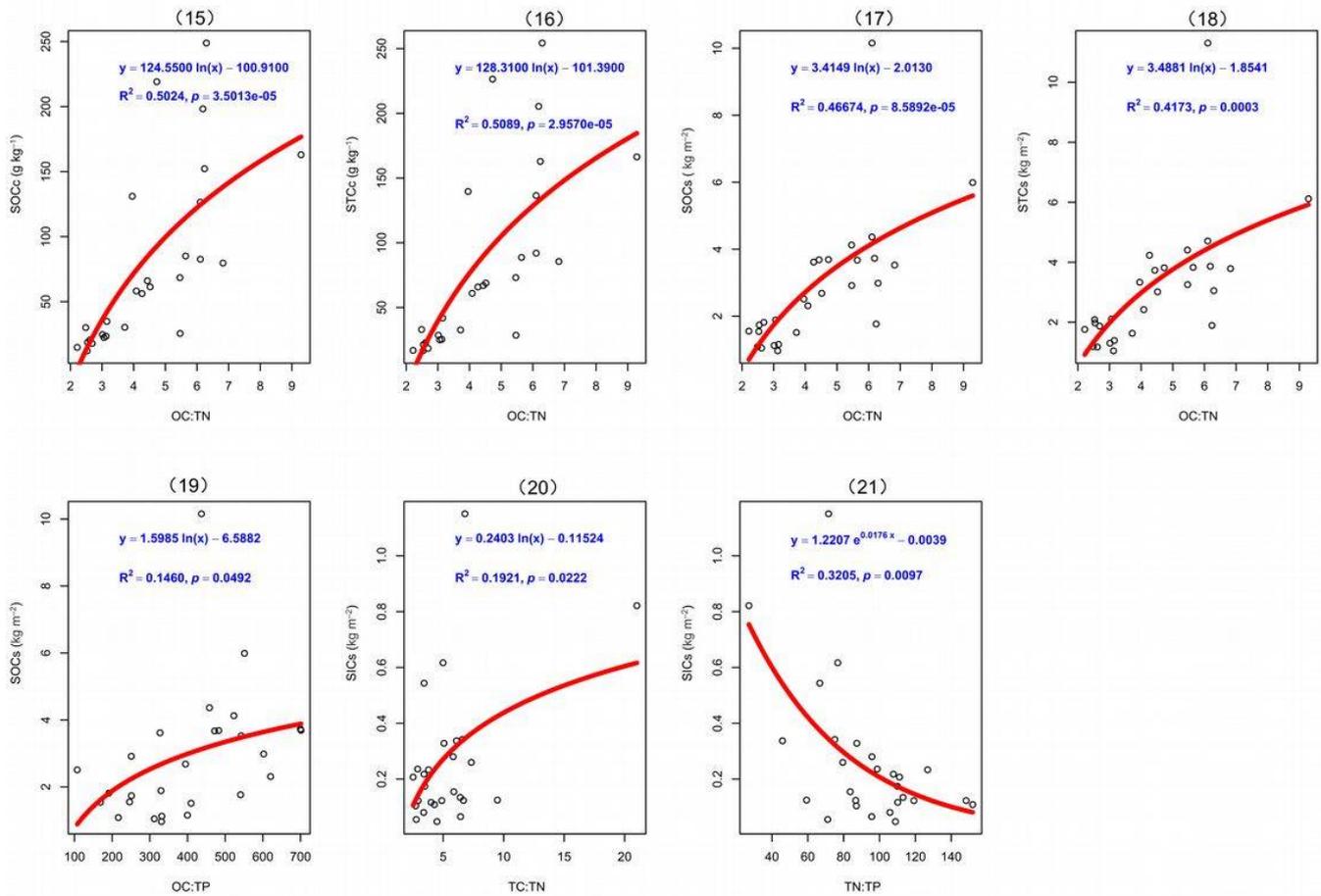
Fig. S2 – (continued).



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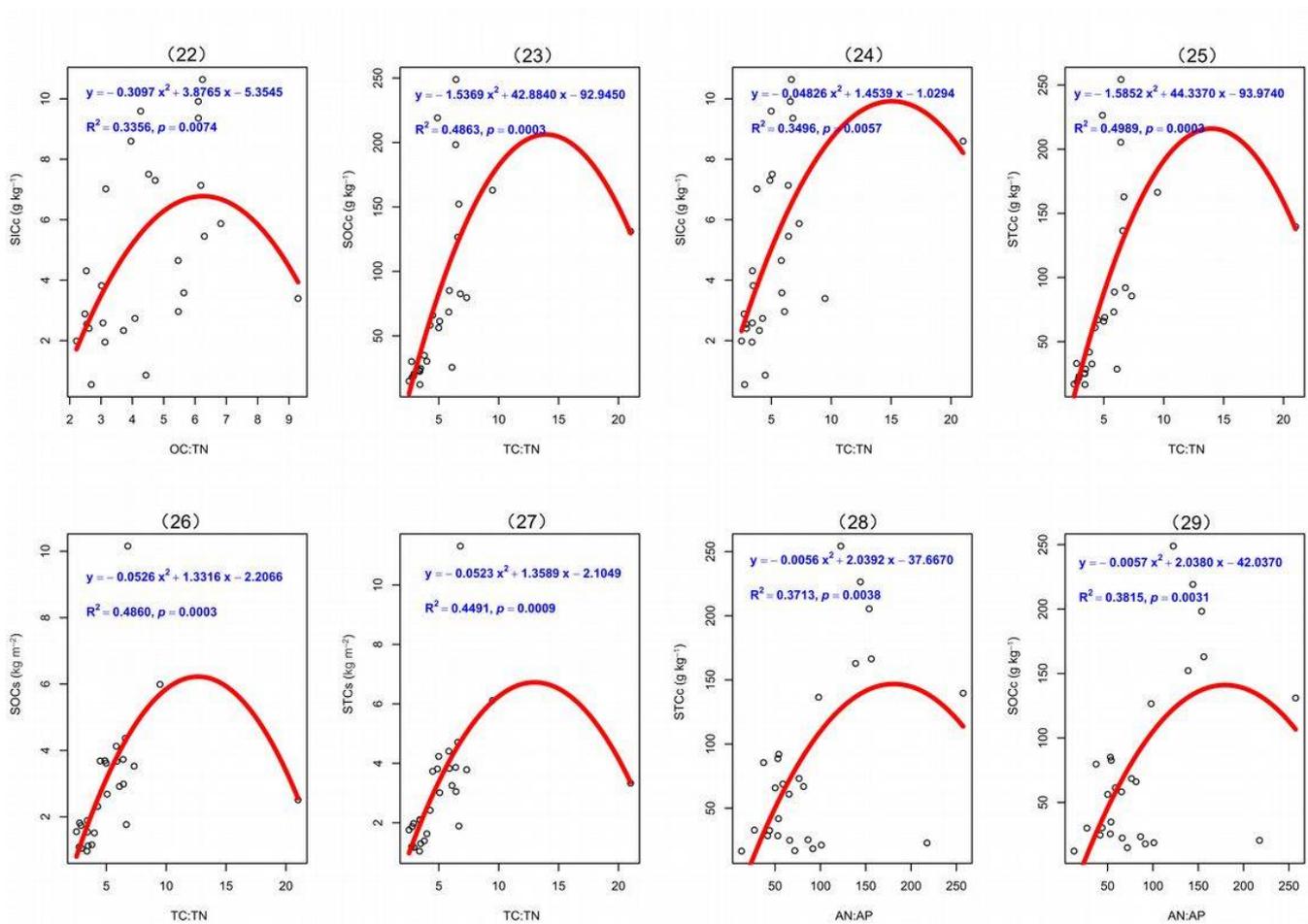
Fig. S2 – (continued).



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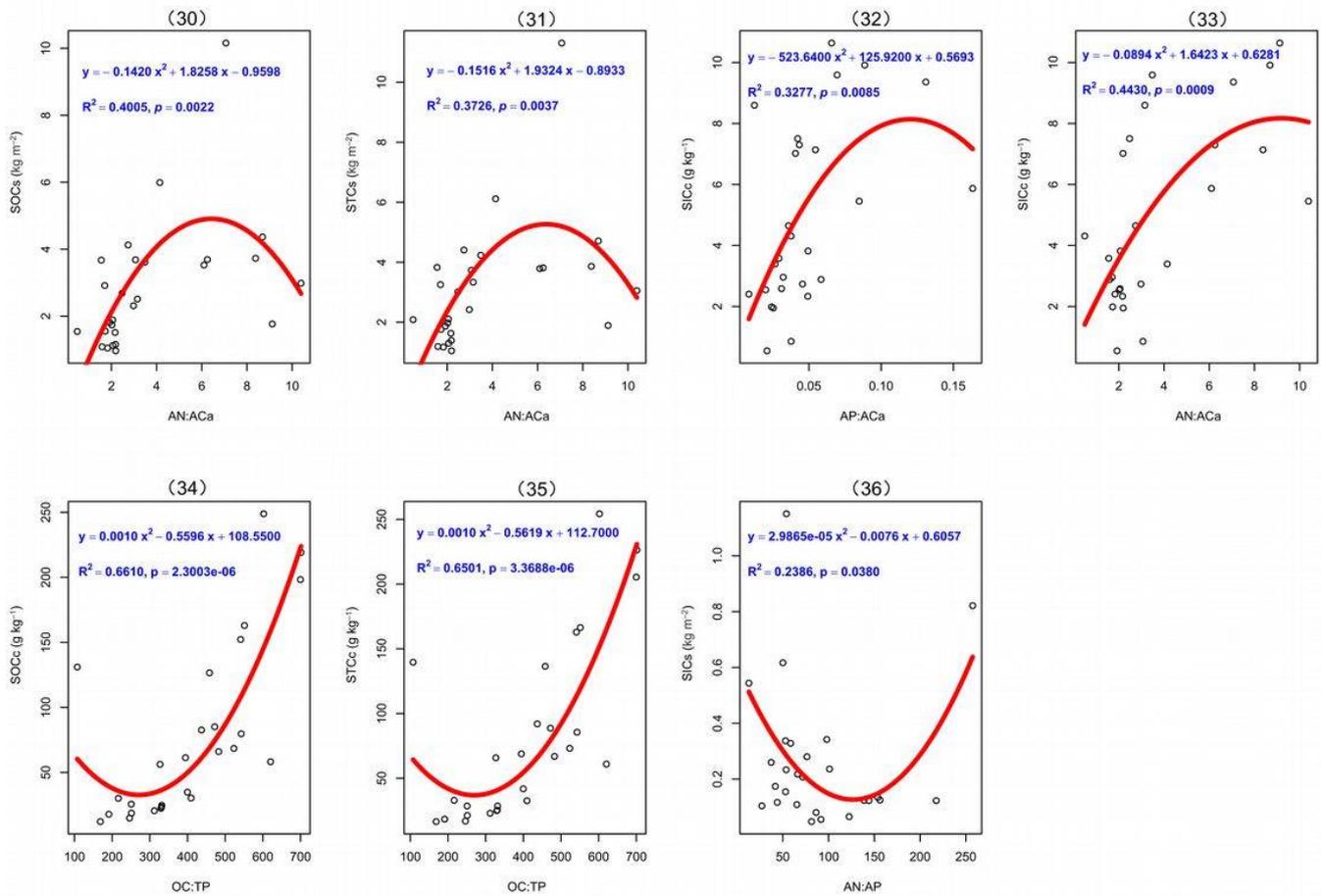
Fig. S2 – (continued).



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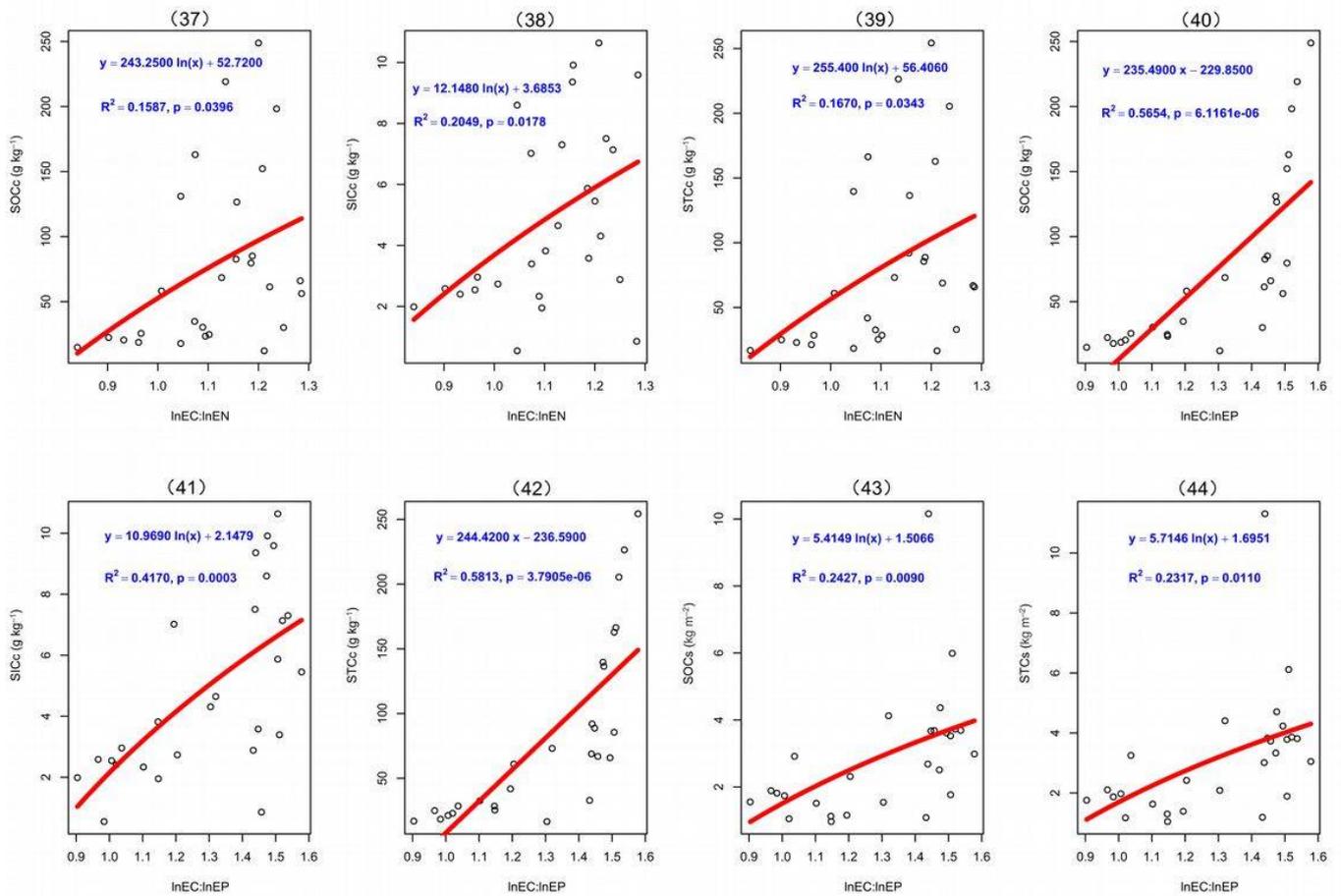
Fig. S2 – (continued).



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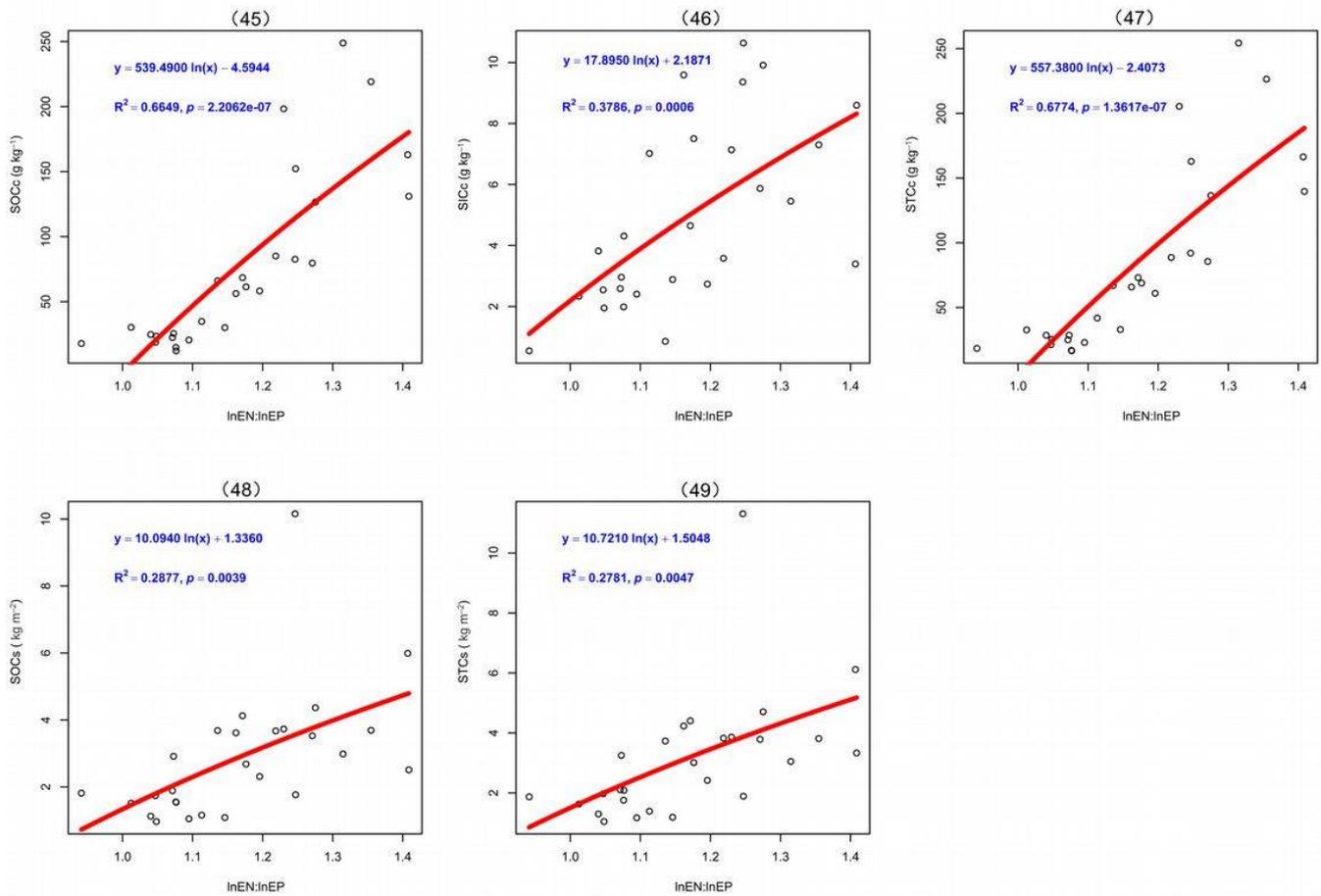
Fig. S2 – (continued).



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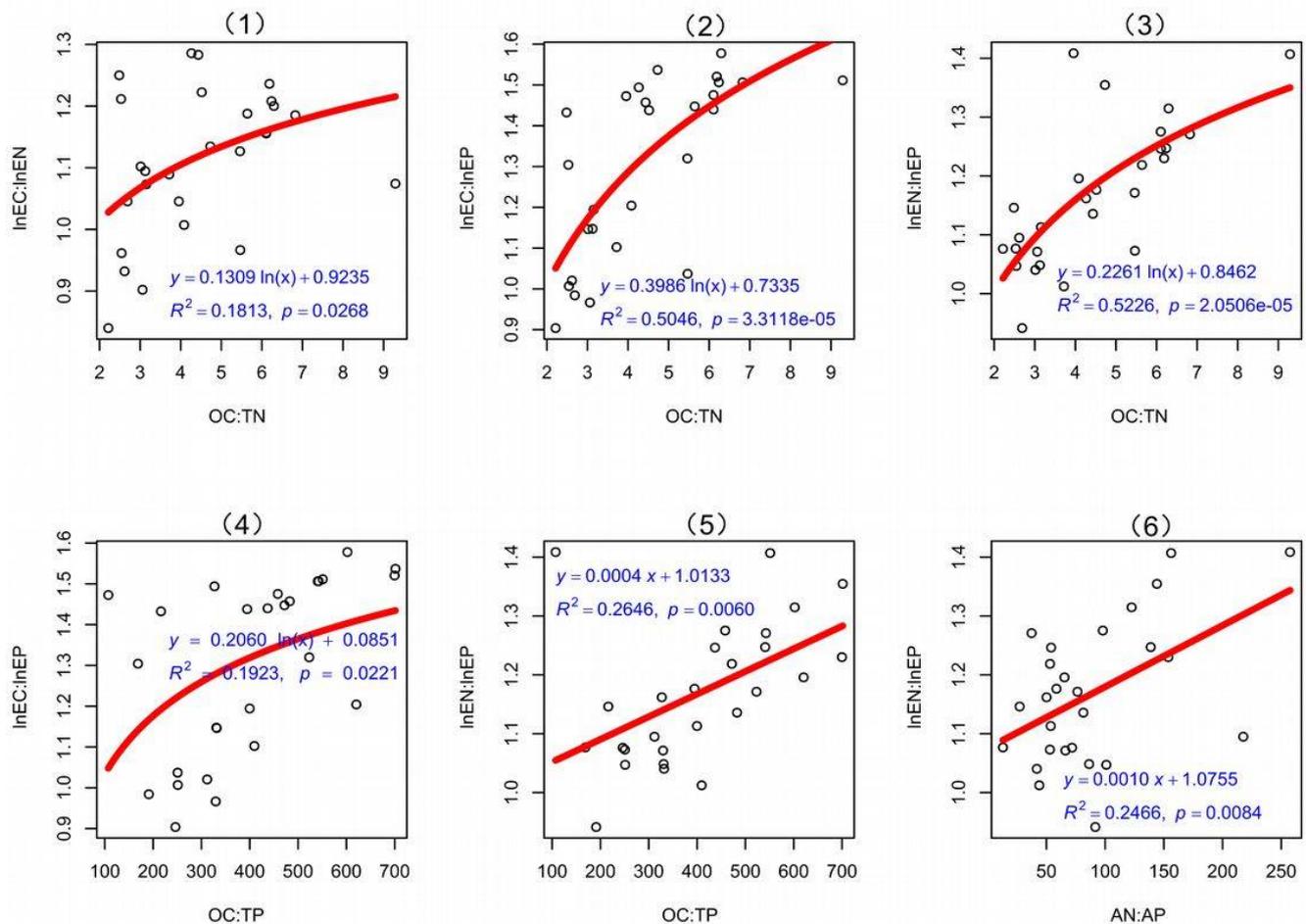
Fig. S2 – (continued).



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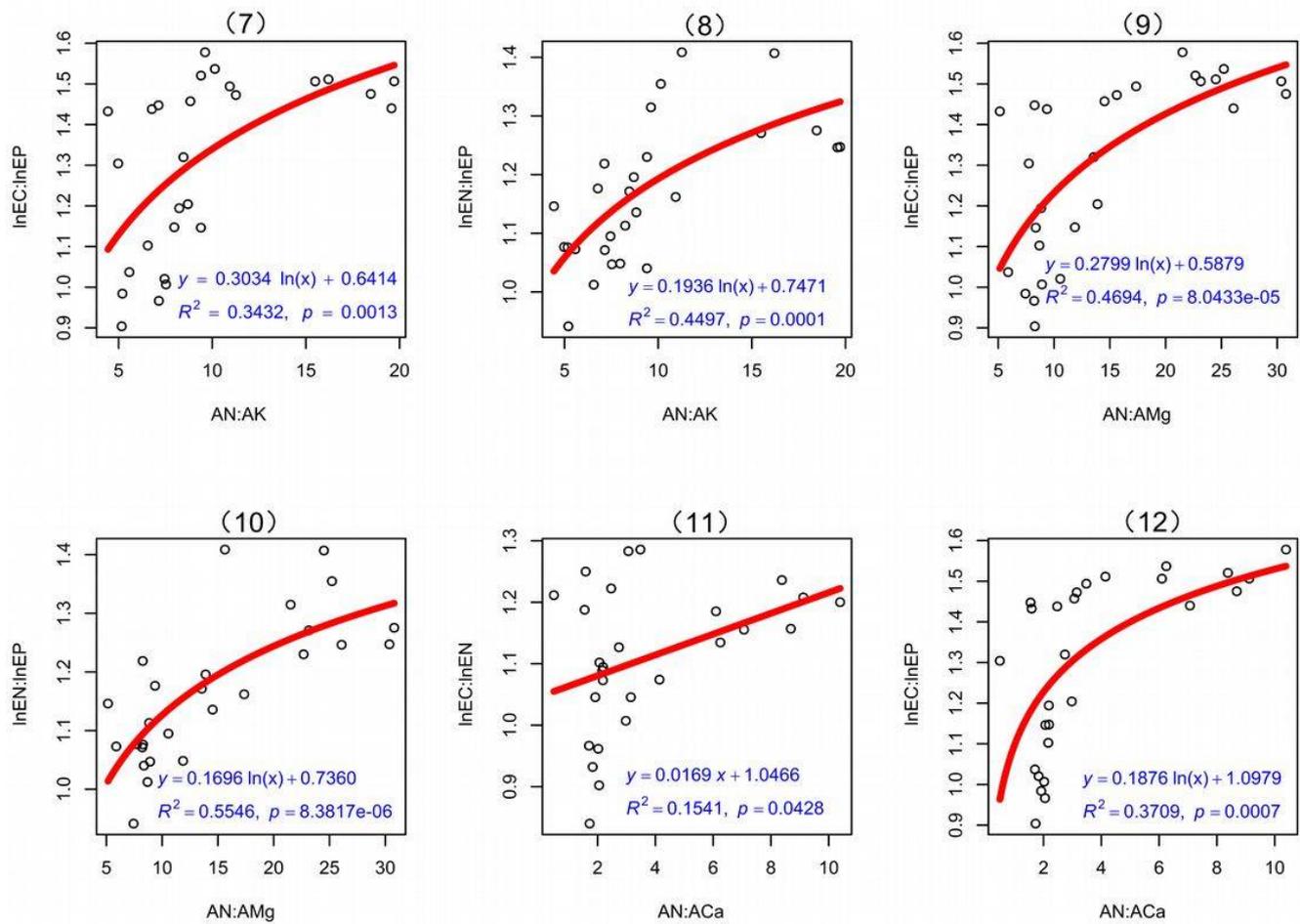
Fig. S3 -Regression analysis reveals soil nutrient stoichiometry ratios increases soil C-, nitrogen (N)- and phosphorus (P)-acquiring enzymes stoichiometry ratios. Abbreviations are listed in the method section of the parent paper.



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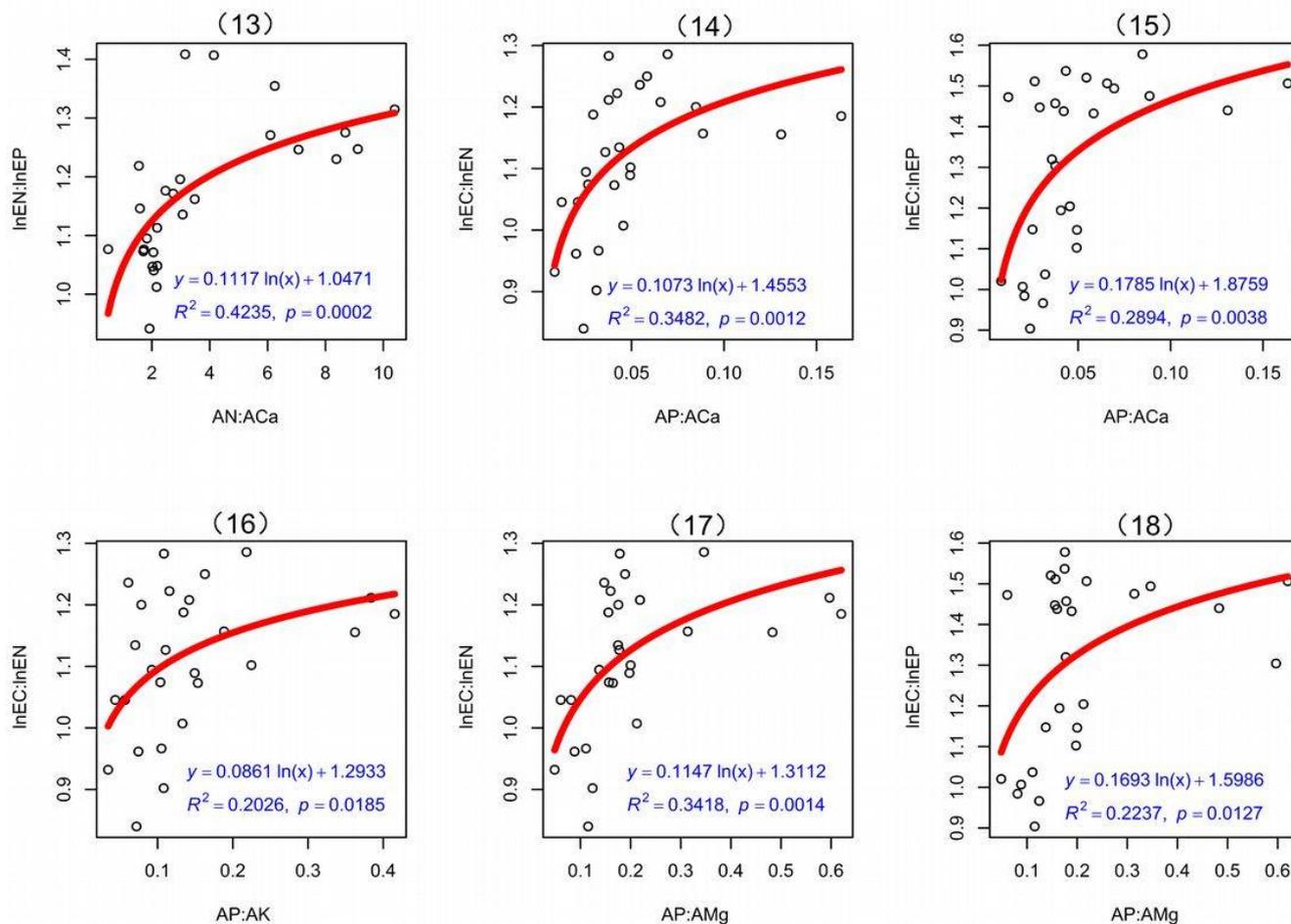
Fig. S3 – (continued).



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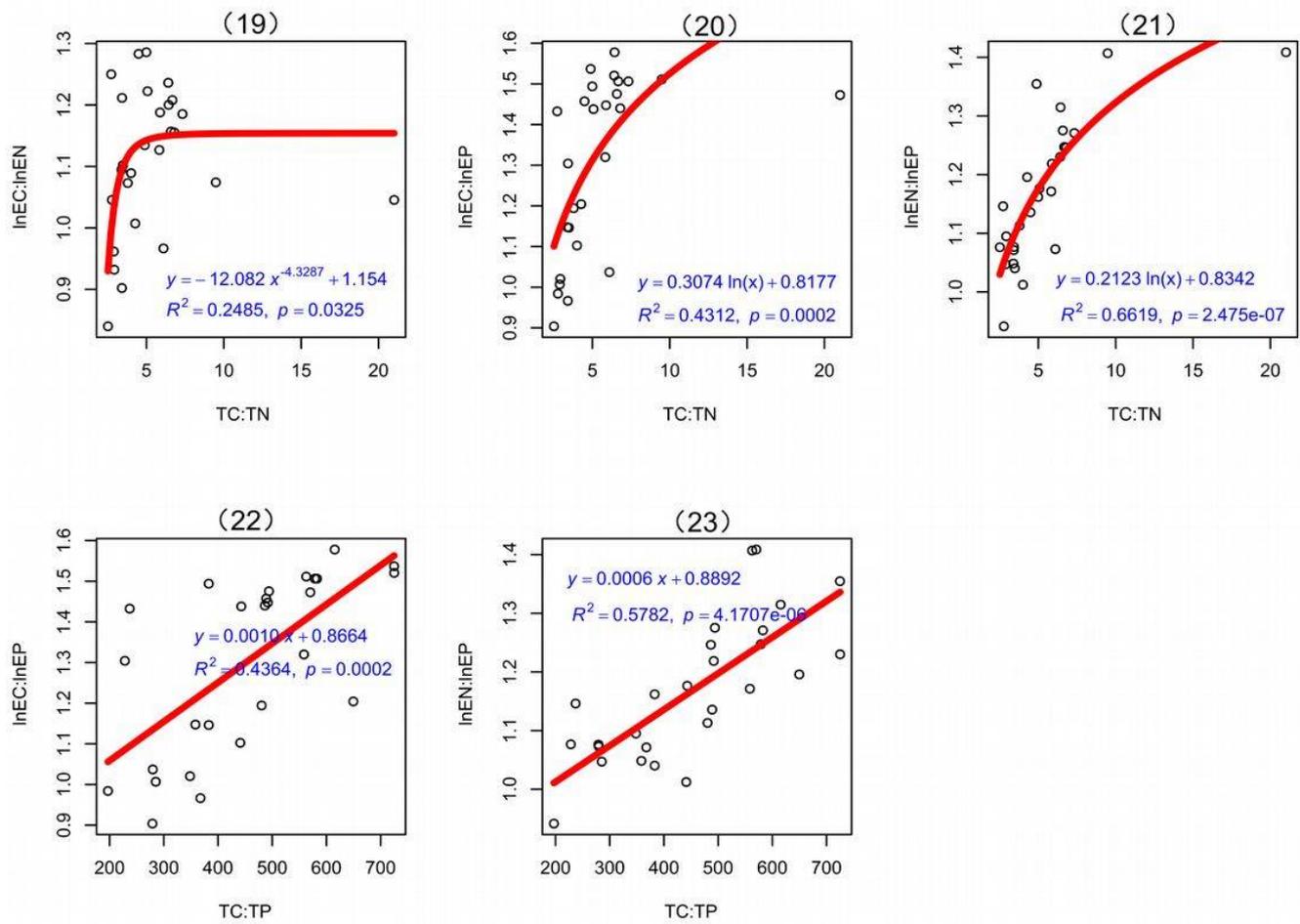
Fig. S3 – (continued).



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Fig. S3 – (continued).



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Fig. S4 - Regression analysis reveals soil inorganic carbon concentration increases with soil organic carbon concentration

